

FILE 'REGISTRY' ENTERED AT 14:20:52 ON 18 APR 2006

=> S RESTRICTION ENZYME/CN

L1 1 RESTRICTION ENZYME/CN

FILE 'CAPLUS' ENTERED AT 14:21:05 ON 18 APR 2006

=> S RESTRICTION ENZYME;S L1;S L2,L3

101497 RESTRICTION

13271 RESTRICTIONS

113935 RESTRICTION

(RESTRICTION OR RESTRICTIONS)

768855 ENZYME

444988 ENZYMES

972576 ENZYME

(ENZYME OR ENZYMES)

L2 16020 RESTRICTION ENZYME

(RESTRICTION(W) ENZYME)

L3 4021 L1

L4 18573 (L2 OR L3)

=> S HELICOBACTER;S PYLORI

11962 HELICOBACTER

52 HELICOBACTERS

L5 11962 HELICOBACTER

(HELICOBACTER OR HELICOBACTERS)

11741 PYLORI

20 PYLORIS

L6 11754 PYLORI

(PYLORI OR PYLORIS)

=> S L5(W)L6

L7 11372 L5(W)L6

=> S L5 AND L4;S L7 AND L4

L8 93 L5 AND L4

L9 83 L7 AND L4

=> S L8,L9

L10 93 (L8 OR L9)

=> S L5 (4A) L4

L11 7 L5 (4A) L4

=> S L5 (6A) L4

L12 9 L5 (6A) L4

=> S L5 (20A) L4

L13 20 L5 (20A) L4

=> S L8 NOT L13

L14 73 L8 NOT L13

=> D L13 1-20 CBIB ABS;D L14 1-73 TI

L13 ANSWER 1 OF 20 CAPLUS COPYRIGHT 2006 ACS on STN

2005:735189 Document No. 143:207265 Methods for mapping of antibiotic resistance gene mutations in whole genomes using restriction enzyme modulation of transformation efficiency. Beutel, Bruce A.; Lerner, Claude G.; Kakavas, Stephan J. (USA). U.S. Pat. Appl. Publ. US 2005176019 A1 20050811, 30 pp. (English). CODEN: USXXCO. APPLICATION: US 2004-775409 20040210.

AB This invention provides methods for mapping of antibiotic resistance gene mutations in whole genomes using restriction enzyme modulation of transformation efficiency. The method comprises isolating genomic DNA from an organism having a mutant phenotype, digesting dna with restriction enzymes and transforming a non mutant host strain with the digested DNA fragments. The frequency with which the host strain is transformed to acquire the mutant phenotype is determined. The location of the mutation is identified by determining the regions of the genome restriction site map, derived from available genomic sequence data that best fit transformation frequency data. These methods may also be used to map mutations in genes associated with increased production of proteins, secondary metabolites, drugs, and improved degradation of waste.

L13 ANSWER 2 OF 20 CAPLUS COPYRIGHT 2006 ACS on STN

2005:575681 Document No. 144:64833 Polymerase chain reaction-restriction fragment length polymorphism analysis in clarithromycin-resistant *Helicobacter pylori* infection in children using stool sample. Booka, Mina; Okuda, Masumi; Shin, Kouichirou; Miyashiro, Eikichi; Hayashi, Hiroko; Yamauchi, Koji; Tamura, Yoshitaka; Yoshikawa, Norishige (Department of Pediatrics, Wakayama Medical University, Wakayama, Japan). *Helicobacter*, 10(3), 205-213 (English) 2005. CODEN: HELIFL. ISSN: 1083-4389. Publisher: Blackwell Publishing Ltd..

AB To analyze clarithromycin-resistant *Helicobacter pylori* infection in children, we developed a method of polymerase chain reaction-restriction fragment length polymorphism (PCR-RFLP) anal. using stool samples. Twenty-three children without significant upper abdominal symptoms were included (mean age 7.0 years). Of these, 18 and five were diagnosed as *H. pylori*-pos. and -neg., resp., by the *H. pylori* stool antigen test (HpSA). The DNA from the stool samples was purified using the QIAamp DNA Stool Minikit (QIAGEN). The PCR was performed on the purified DNA using oligonucleotide primers designed to amplify the 23S rRNA gene of *H. pylori*. The PCR products were reacted with restriction enzymes MboII, BceAI, and BsaI to detect mutations A2142G, A2142C, and A2143G, resp. Sixteen of the 18 HpSA-pos. samples were PCR-pos., and all five HpSA-neg. samples were PCR-neg. Thus, the PCR had 89% sensitivity and 100% specificity, with 91% accuracy in reference to HpSA. Of the 16 PCR-pos. samples, one and four were digested with MboII and BsaI, resp., indicating 31% prevalence of CAM-resistance. We conclude that the PCR-RFLP using stool samples is a rapid and reliable method to noninvasively detect clarithromycin-resistant *H. pylori* infection in children. It may be useful before choosing regimens of *H. pylori* eradication.

L13 ANSWER 3 OF 20 CAPLUS COPYRIGHT 2006 ACS on STN

2005:222941 Document No. 143:225290 Use of the restriction enzyme EcoRI for pulsed-field gel electrophoretic analysis of *Helicobacter pylori*. Hosaka, Yoshio; Irinoda, Kazuhiko; Nakano, Ryuichi; Tanabe, Satoshi; Koizumi, Wasaburo; Saigenji, Katunori; Inoue, Matsuhisa (Department of Microbiology, First Department of Internal Medicine, Kitasato University School of Medicine, Kanagawa, Japan). *Journal of Clinical Microbiology*, 43(2), 931-932 (English) 2005. CODEN: JCMIDW. ISSN: 0095-1137. Publisher: American Society for Microbiology.

AB Pulsed-field gel electrophoretic (PFGE) anal. of *Helicobacter pylori* isolates is not commonly employed because of the inability to compare the typing with other typing systems. The authors adapted the PFGE anal. for *H. pylori* by EcoRI and slightly modified their laboratory methods to improve the typing of isolates (type-ability was 97%).

L13 ANSWER 4 OF 20 CAPLUS COPYRIGHT 2006 ACS on STN

2003:341381 Document No. 139:31393 Use of polymerase chain reaction and enzymatic cleavage in the identification of *Helicobacter* spp. in gastric mucosa of human beings from North Parana, Brazil. Camargo, P. L.; Alfieri, A. A.; Bracarense, A. P. F. R. L.; Menoli, R.; Spinosa, S. R.; Hagiwara, M. K. (Departamento de Clinicas Veterinarias, Faculdade de Medicina, Universidade Estadual de Londrina, Londrina, PR, 86051-970, Brazil). *Memorias do Instituto Oswaldo Cruz*, 98(2), 265-268 (English) 2003. CODEN: MIOCAS. ISSN: 0074-0276. Publisher: Instituto Oswaldo Cruz.

AB *Helicobacter pylori* is the most common gastric bacteria of human beings. Animal-borne helicobacter have been associated with gastritis, ulceration, and gastric mucosa-associated lymphoid-tissue lymphoma in people. We attempted to identify the species of *Helicobacter* spp. that infect human beings in north Parana, Brazil. Samples of gastric mucosa from 38 dyspeptic patients were analyzed by optic microscopy on silver stained slides, polymerase chain reaction (PCR), and enzymic cleavage. Genus and species-specific primers to *H. pylori*, *H. heilmannii*, *H. felis*, and consensual primers to *H. bizzozeronii* or *H. salomonis* were used. The PCR products were submitted to enzymic cleavage by *VspI* (*Helicobacter* spp. product) and *HinfI* (species products) enzymes. Thirty-two out of 38 patients evaluated had 3.2 to 5 μ m long bacteria that resembled *H. pylori* in Warthin-Starry stained slides and were pos. to the genus *Helicobacter* by PCR. In 30 of these patients the bacteria were identified as *H. pylori*. Two samples pos. by silver stain were neg. to all species tested by PCR. None of the 38 samples was pos. to animal-origin helicobacter species. These results show that PCR and enzymic restriction are practical methods to identify the species of helicobacters present in gastric mucosa of human beings. People in north Parana appear to be infected mostly with *H. pylori*.

L13 ANSWER 5 OF 20 CAPLUS COPYRIGHT 2006 ACS on STN

2003:52021 Document No. 138:317350 *Helicobacter pylori* interstrain restriction-modification diversity prevents genome subversion by chromosomal DNA from competing strains. Aras, Rahul A.; Small, Aaron J.; Ando, Takafumi; Blaser, Martin J. (Department of Medicine and Microbiology, New York University School of Medicine and VA Medical Center, New York, NY, USA). *Nucleic Acids Research*, 30(24), 5391-5397 (English) 2002. CODEN: NARHAD. ISSN: 0305-1048. Publisher: Oxford University Press.

AB *Helicobacter pylori*, bacteria that colonize the human gastric mucosa, possess a large number of genes for restriction-modification (R-M) systems, and essentially, every strain possesses a unique complement of functional and partial R-M systems. Nearly half of the *H. pylori* strains studied possess an active type IIs R-M system, *HpyII*, with the recognition sequence GAAGA. Recombination between direct repeats that flank the R-M cassette allows for its deletion whereas strains lacking *hpyIIRM* can acquire this cassette through natural transformation. We asked whether strains lacking *HpyII* R-M activity can acquire an active *hpyIIRM* cassette [containing a 1.4 kb kanamycin resistance (*aphA*) marker], whether such acquisition is DNase sensitive or resistant and whether restriction barriers limit acquisition of chromosomal DNA. Our results indicate that natural transformation and conjugation-like mechanisms may contribute to the transfer of large (4.8 kb) insertions of chromosomal DNA between *H. pylori* strains, that inactive or partial R-M systems can be reactivated upon recombination with a functional allele, consistent with their being contingency genes, and that *H. pylori* R-M diversity limits acquisition of chromosomal DNA fragments of ≥ 1 kb.

L13 ANSWER 6 OF 20 CAPLUS COPYRIGHT 2006 ACS on STN

2002:923768 Document No. 138:199687 Identification of strain-specific genes located outside the plasticity zone in nine clinical isolates of *Helicobacter pylori*. Chanto, Grettel; Occhialini, Alessandra; Gras,

Nathalie; Alm, Richard A.; Megraud, Francis; Marais, Armelle (Laboratoire de Bacteriologie, Universite Victor Segalen Bordeaux 2 and Hopital Pellegrin, Bordeaux, 33076, Fr.). Microbiology (Reading, United Kingdom), 148(11), 3671-3680 (English) 2002. CODEN: MROBEO. ISSN: 1350-0872. Publisher: Society for General Microbiology.

- AB Helicobacter pylori is a Gram-neg. bacterium that is associated with the development of peptic ulcers and gastric carcinoma in humans. This species appears to be one of the most genetically variable bacteria described to date. The overall level of heterogeneity within strains of this organism was determined by comparing the genome sequences of two reference strains, J99 and 26695. The aim of this study was to measure the genetic diversity within strains of H. pylori by looking for strain-specific genes in nine H. pylori strains isolated from patients suffering from chronic gastritis (n=3), duodenal ulcers (n=3) or gastric cancer (n=3). Seven loci that contained strain-specific genes in strains J99 and 26695 were studied. These regions were subsequently amplified from most of the clin. isolates studied and their sequences were determined. ORFs were predicted from the sequence data and were compared to sequences within the databases. The results showed that the genes flanking the ORFs specific to either strain J99 or strain 26695 were also present in a similar configuration in the genomes of the nine clin. isolates. Moreover, in most regions, ORFs homologous to those found in the corresponding loci in the two reference strains were detected. However, in 10 regions, genes similar to those located at another locus in the genome of J99 or 26695 were found. Finally, six strain-specific genes were identified in three regions of three of the H. pylori strains isolated from patients with duodenal ulcers (n=2) and gastric cancer (n=1). Of these six genes, five were putative genes and one was an orthologue of a gene encoding a transposase in Thermotoga maritima. However, no association with disease was found for these genes.

L13 ANSWER 7 OF 20 CAPLUS COPYRIGHT 2006 ACS on STN

2002:897447 Document No. 138:249565 Transcriptional phase variation of a type III restriction-modification system in Helicobacter pylori. De Vries, Nicolette; Duinsbergen, Dirk; Kuipers, Ernst J.; Pot, Raymond G. J.; Wiesenekker, Patricia; Penn, Charles W.; Van Vliet, Arnoud H. M.; Vandenbroucke-Grauls, Christina M. J. E.; Kusters, Johannes G. (Department of Gastroenterology and Hepatology, Erasmus MC-University Medical Center Rotterdam, Rotterdam, 3015 GD, Neth.). Journal of Bacteriology, 184(23), 6615-6623 (English) 2002. CODEN: JOBAAY. ISSN: 0021-9193. Publisher: American Society for Microbiology.

- AB Phase variation is important in bacterial pathogenesis, since it generates antigenic variation for the evasion of immune responses and provides a strategy for quick adaptation to environmental changes. In this study, a Helicobacter pylori clone, designated MOD525, was identified that displayed phase-variable lacZ expression. The clone contained a transcriptional lacZ fusion in a putative type III DNA methyltransferase gene (mod, a homolog of the gene JHP1296 of strain J99), organized in an operon-like structure with a putative type III restriction endonuclease gene (res, a homolog of the gene JHP1297), located directly upstream of it. This putative type III restriction-modification system was common in H. pylori, as it was present in 15 out of 16 clin. isolates. Phase variation of the mod gene occurred at the transcriptional level both in clone MOD525 and in the parental H. pylori strain 1061. Further anal. showed that the res gene also displayed transcriptional phase variation and that it was cotranscribed with the mod gene. A homopolymeric cytosine tract (C tract) was present in the 5' coding region of the res gene. Length variation of this C tract caused the res open reading frame (ORF) to shift in and out of frame, switching the res gene on and off at the translational level. Surprisingly, the presence of an intact res ORF was pos. correlated with active transcription of the downstream mod gene. Moreover, the C tract was required for the occurrence of transcriptional phase variation. Our finding that translation and transcription are linked during phase variation through slipped-strand mispairing is new for H. pylori.

L13 ANSWER 8 OF 20 CAPLUS COPYRIGHT 2006 ACS on STN

2001:811460 Document No. 136:82482 Regulation of the HpyII restriction-modification system of *Helicobacter pylori* by gene deletion and horizontal reconstitution. Aras, Rahul A.; Takata, Tohru; Ando, Takafumi; Van der Ende, Arie; Blaser, Martin J. (Departments of Medicine and Microbiology, New York University School of Medicine and VA Medical Center, New York, NY, USA). *Molecular Microbiology*, 42(2), 369-382 (English) 2001. CODEN: MOMIEE. ISSN: 0950-382X. Publisher: Blackwell Science Ltd..

AB *H. pylori*, Gram-neg., curved bacteria colonizing the human stomach, possess strain-specific complements of functional restriction-modification (R-M) systems. Restriction-modification systems have been identified in most bacterial species studied and are believed to have evolved to protect the host genome from invasion by foreign DNA. The large number of R-Ms homologous to those in other bacterial species and their strain-specificity suggest that *H. pylori* may have horizontally acquired these genes. A type IIS restriction-modification system, hpyIIRM, was active in 2 of the 6 *H. pylori* strains studied. We demonstrate now that in most strains lacking M.HpyII function, there is complete absence of the R-M system. Direct DNA repeats of 80 bp flanking the hpyIIRM system allow its deletion, resulting in an "empty-site" genotype. Strains possessing this empty-site genotype and strains with a full but inactive hpyIIRM can reacquire the hpyIIRM cassette and functional activity through natural transformation by DNA from the parental R-M+ strain. Identical isolates divergent for the presence of an active HpyII R-M pose different restriction barriers to transformation by foreign DNA. That *H. pylori* can lose HpyII R-M function through deletion or mutation, and can horizontally reacquire the hpyIIRM cassette, is, in composite, a novel mechanism for R-M regulation, supporting the general hypothesis that *H. pylori* populations use mutation and transformation to regulate gene function.

L13 ANSWER 9 OF 20 CAPLUS COPYRIGHT 2006 ACS on STN

2000:690153 Document No. 133:346932 Overcoming the restriction barrier to plasmid transformation of *Helicobacter pylori*. Donahue, John P.; Israel, Dawn A.; Peek, Richard M., Jr.; Blaser, Martin J.; Miller, Geraldine G. (Department of Medicine, Vanderbilt University School of Medicine, Nashville, TN, 37232, USA). *Molecular Microbiology*, 37(5), 1066-1074 (English) 2000. CODEN: MOMIEE. ISSN: 0950-382X. Publisher: Blackwell Science Ltd..

AB *H. pylori* strains demonstrate substantial variability in the efficiency of transformation by plasmids from *Escherichia coli*, and many strains are completely resistant to transformation. Among the barriers to transformation are numerous strain-specific restriction-modification systems in *H. pylori*. A method was developed to protect plasmid DNA from restriction by *in vitro* site-specific methylation using cell-free exts. of *H. pylori* before transformation. In 2 cases, plasmid DNA treated with cell-free exts. *in vitro* acquired the restriction pattern characteristic of genomic DNA from the source strain. Among 3 strains examined in detail, the transformation frequency by treated plasmid shuttle and suicide vectors was significantly increased compared with mock-treated plasmid DNA. The results indicate that the restriction barrier in *H. pylori* can be largely overcome by specific DNA methylation *in vitro*. The approach described should significantly enhance the ability to manipulate gene function in *H. pylori* and other organisms that have substantial restriction barriers to transformation.

L13 ANSWER 10 OF 20 CAPLUS COPYRIGHT 2006 ACS on STN

2000:690152 Document No. 133:346931 Restriction-modification system differences in *Helicobacter pylori* are a barrier to interstrain plasmid transfer. Ando, Takafumi; Xu, Qing; Torres, Melaine; Kusugami, Kazuo; Israel, Dawn A.; Blaser, Martin J. (Division of Infectious Diseases, Department of Medicine, Vanderbilt University School of Medicine and VA Medical Center, Nashville, TN, USA). *Molecular Microbiology*, 37(5), 1052-1065 (English) 2000. CODEN: MOMIEE. ISSN: 0950-382X. Publisher: Blackwell Science Ltd..

AB H. pylori cells are naturally competent for the uptake of both plasmid and chromosomal DNA. However, there are strong barriers to transformation of H. pylori strains by plasmids derived from unrelated strains. The mol. mechanisms underlying these barriers were studied. Transformation efficiency was assessed using pHP1, an Escherichia coli-H. pylori shuttle vector conferring kanamycin resistance. Transformation of 33 H. pylori strains was attempted with pHP1 purified from either E. coli or H. pylori, and was successfully introduced into only 11 strains. Digestion of H. pylori chromosomes with different restriction endonucleases (REs) showed that DNA methylation patterns vary substantially among strains. The strain most easily transformed, JP26, has extremely low endogenous RE activity and lacks a restriction-modification (R-M) system, homologous to MboI, which is highly conserved among H. pylori strains. When this system was introduced to JP26, pHP1 from MboI.M+ JP26, but not from wild-type JP26, transformed MboI R-M+ JP26 and heterologous MboI R-M+ wild-type H. pylori strains. Parallel studies with pHP1 from dam+ and dam- E. coli strains confirmed these findings. These data indicate that the endogenous REs of H. pylori strains represent a critical barrier to interstrain plasmid transfer among H. pylori.

L13 ANSWER 11 OF 20 CAPLUS COPYRIGHT 2006 ACS on STN
2000:482587 Document No. 133:85884 Molecular analysis of restriction-modification systems in Helicobacter pylori. Xu, Quing (Vanderbilt University, USA). 156 pp. Avail. UMI, Order No. DA9944589 From: Diss. Abstr. Int., B 2000, 60(9), 4433 (English) 1999.
AB Unavailable

L13 ANSWER 12 OF 20 CAPLUS COPYRIGHT 2006 ACS on STN
2000:394546 Document No. 133:277046 Purification of the novel endonuclease, Hpy188I, and cloning of its restriction-modification genes reveal evidence of its horizontal transfer to the Helicobacter pylori genome. Xu, Qing; Stickel, Shawn; Roberts, Richard J.; Blaser, Martin J.; Morgan, Richard D. (Department of Microbiology and Immunology, Vanderbilt University School of Medicine, Nashville, TN, 37232, USA). Journal of Biological Chemistry, 275(22), 17086-17093 (English) 2000. CODEN: JBCHA3. ISSN: 0021-9258. Publisher: American Society for Biochemistry and Molecular Biology.

AB We have isolated a novel restriction endonuclease, Hpy188I, from Helicobacter pylori strain J188. Hpy188I recognizes the unique sequence, TCNGA, and cleaves the DNA between nucleotides N and G in its recognition sequence to generate a one-base 3' overhang. Cloning and sequence anal. of the Hpy188I modification gene in strain J188 reveal that hpy188IM has a 1299-base pair (bp) open reading frame (ORF) encoding a 432-amino acid product. The predicted protein sequence of Hpy188I contains conserved motifs typical of aminomethyltransferases, and Western blotting indicates that it is an N-6 adenine methyltransferase. Downstream of hpy188IM is a 513-bp ORF encoding a 170-amino acid product, that has a 41-bp overlap with hpy188IM. The predicted protein sequence from this ORF matches the amino acid sequence obtained from purified Hpy188I, indicating that it encodes the endonuclease. The Hpy188I R-M genes are not present in either strain of H. pylori that has been completely sequenced but are found in two of 11 H. pylori strains tested. The significantly lower G + C content of the Hpy188I R-M genes implies that they have been introduced relatively recently during the evolution of the H. pylori genome.

L13 ANSWER 13 OF 20 CAPLUS COPYRIGHT 2006 ACS on STN
2000:267521 Document No. 132:305394 Genome analysis of Helicobacter pylori by pulsed-field gel electrophoresis. Chen, Gelin; Fukuchi, Kunihiro; Sakurai, Koichi; Hattori, Takashi; Ito, Ribu; Wakuta, Rika; Takagi, Yasushi; Fujita, Rikiya; Gomi, Kunihide (Dep. Clin. Pathol., Sch. Med., Showa Univ., Shinagawa-ku, Tokyo, 142-8666, Japan). Rinsho Byori, 48(4), 348-354 (Japanese) 2000. CODEN: RBYOAI. ISSN: 0047-1860. Publisher: Nippon Rinsho Byori Gakkai.

AB The mol. epidemiol. of total 121 isolates of *Helicobacter pylori* was analyzed by pulsed-field gel electrophoresis method with restriction enzyme Spe I. Seventy-seven isolates were separated from the clin. samples, 36 isolates from pyloric antrum and the body of stomach of 18 patients and 8 isolates from pyloric antrum of 4 patients that include one colony before and after sterilizing treatment to each patient. Seventy-five in 77 isolates showed different genomic types resp., and the other 2 isolates had the same genomic type and were suspected to be caused by cross infection of medical workers or the instruments that used in examination because they were from patients who were examined by gastric microscope in same time and same laboratory. In isolates from 4 patients who were treated by sterilizing method, 2 patients showed same genomic types with that observed before the treatment, and one patient showed an incomplete treatment because the genomic type of its colony was is similar, and another patient could be infected again because its isolates showed different genomic type. In 18 patients whose isolates were separated from pyloric antrum and body of the stomach resp. to each person, isolates of 3 patients showed different genomic types in the two different part of stomach indicating that they had two and more clones of *H. pylori*.

L13 ANSWER 14 OF 20 CAPLUS COPYRIGHT 2006 ACS on STN
1999:582413 Document No. 132:9499 Differences among *Helicobacter pylori* strains isolated from three different populations and demonstrated by restriction enzyme analysis of an internal fragment of the conserved gene hpaA. Evans, Dolores G.; Queiroz, Dulciene M. M.; Mendes, Edilberto N.; Svennerholm, Ann-Marie; Evans, Doyle J., Jr. (Department of Medicine, Baylor College of Medicine, and Bacterial, Veterans Affairs Medical Center, Houston, TX, 77030, USA). *Helicobacter*, 4(2), 82-88 (English) 1999. CODEN: HELIFL. ISSN: 1083-4389. Publisher: Blackwell Science.

AB Our goal was to test the idea that *Helicobacter pylori* genotypes vary from one population to another. Anal. of Sau3A and HinfI restriction fragment-length polymorphism (RFLP) in a 375-bp polymerase chain reaction amplicon of hpaA was used to compare 31 *H. pylori* isolates from a relatively small and genetically homogeneous population (Goteborg, Sweden) with those of large, genetically heterogeneous populations located in two different countries (50 isolates from Houston, TX, and 69 isolates from Minas Gerais, a state in the southeastern region of Brazil). Five different Sau3A and three different HinfI restriction patterns were found; different combinations of these comprise 10 different RFLP types, I through X. The RFLP types found in the United States and Brazil collections were very similar, except for two Brazil isolates belonging to type VIII and five Brazil isolates belonging to type X, neither type found in the United States. The overall profile of *H. pylori* isolates from Sweden was remarkably different, with 18 of 31 (58%) having a new Sau3A restriction pattern, termed gS; 10 of these 18 isolates had HinfI restriction pattern E (RFLP type VIII), and 8 had HinfI restriction pattern F (RFLP type IX). No isolates from Sweden belonged to RFLP type III or type X. RFLP typing of a 375-bp polymerase chain reaction-amplified DNA fragment of *H. pylori* hpaA revealed that *H. pylori* genotypes can and do vary from one population to another. We conclude that the unique RFLP profile shown by the group of *H. pylori* isolates from Goteborg is the result of a cohort effect in this relatively small, stable, genetically homogeneous population. Also, the overall similarity between RFLP profiles of the *H. pylori* isolates from Texas and Minas Gerais coincides with the fact that although geog. distanced, these populations are similar in being large, dynamic, and genetically heterogeneous.

L13 ANSWER 15 OF 20 CAPLUS COPYRIGHT 2006 ACS on STN
1999:311551 Document No. 131:139992 Molecular methods for typing of *Helicobacter pylori* and their applications. Colding, Hanne; Hartzen, Susanne H.; Roshanisehat, Houmayoun; Andersen, Leif Percival; Krogfelt, Karen Angeliki (Institute of Medical Microbiology and Immunology, University of Copenhagen, Copenhagen, DK-2200, Den.). *FEMS Immunology and*

Medical Microbiology, 24(2), 193-199 (English) 1999. CODEN: FIMIEV.
ISSN: 0928-8244. Publisher: Elsevier Science B.V..

- AB A review with 53 refs. Microbial typing is a useful tool in clin. epidemiol. for defining the source and route of infection, for studying the persistence and reinfection rates, clonal selection in the host and bacterial evolution. Phenotypic methods such as biotyping, serotyping and hemagglutinin typing have little discriminatory power compared to genotypic methods concerning the typing of *Helicobacter pylori*. Therefore great efforts have been made to establish useful mol. typing methods. In this context, the most frequently used genotypic methods are described based on our own experience and the literature: (1) restriction endonuclease anal., (2) endonuclease anal. using pulsed-field gel electrophoresis, (3) ribotyping, (4) polymerase chain reaction (using either random primers or repetitive DNA sequence primers), and (5) polymerase chain reaction-restriction fragment length polymorphism anal. of e.g. the urease genes. Furthermore, reproducibility, discriminatory power, ease of performance and interpretation, cost and toxic procedures of each method are assessed. To date no direct comparison of all the mol. typing methods described has been performed in the same study with the same *H. pylori* strains. However, PCR anal. of the urease gene directly on suspensions of *H. pylori* or gastric biopsy material seems to be useful for routine use and applicable in specific epidemiol. situations.

L13 ANSWER 16 OF 20 CAPLUS COPYRIGHT 2006 ACS on STN

1998:249973 Document No. 129:39574 Analyses of the cag pathogenicity island of *Helicobacter pylori*. Akopyants, Natalia S.; Clifton, Sandra W.; Kersulyte, Dangeruta; Crabtree, Jean E.; Youree, Bryan E.; Reece, C. Adonis; Bukanov, Nick O.; Drazek, E. Susan; Roe, Bruce A.; Berg, Douglas E. (Department of Molecular Microbiology, Washington University School of Medicine, St. Louis, MO, 63110, USA). Molecular Microbiology, 28(1), 37-53 (English) 1998. CODEN: MOMIEE. ISSN: 0950-382X. Publisher: Blackwell Science Ltd..

- AB Most strains of *Helicobacter pylori* from patients with peptic ulcer disease or intestinal-type gastric cancer carry cagA, a gene that encodes an immunodominant protein of unknown function, whereas many of the strains from asymptotically infected persons lack this gene. Recent studies showed that the cagA gene lies near the right end of a ~37 kb DNA segment (a pathogenicity island, or PAI) that is unique to cagA+ strains and that the cagPAI was split in half by a transposable element insertion in the reference strain NCTC11638. In complementary expts. reported here, we also found the same cagPAI, and sequenced a 39 kb cosmid clone containing the left "cagII" half of this PAI. Encoded in cagII were four proteins each with homol. to four components of multiprotein complexes of *Bordetella pertussis* ("Ptl"), *Agrobacterium tumefaciens* ("Vir"), and conjugative plasmids ("Tra") that help deliver pertussis toxin and T (tumor inducing) and plasmid DNA, resp., to target eukaryotic or prokaryotic cells, and also homologues of eukaryotic proteins that are involved in cytoskeletal structure. To the left of cagII in this cosmid were genes for homologues of HslU (heat-shock protein) and Era (essential GTPase); to the right of cagII were homologues of genes for a type I restriction endonuclease and ion transport functions. Deletion of the cag PAI had no effect on synthesis of the vacuolating cytotoxin, but this deletion and several cag insertion mutations blocked induction of synthesis of proinflammatory cytokine IL-8 in gastric epithelial cells. Comparisons among *H. pylori* strains indicated that cag PAI gene content and arrangement are rather well conserved. We also identified two genome rearrangements with end-points in the cag PAI. One, in reference strain NCTC11638, involved IS605, a recently described transposable element (as also found by others). Another rearrangement, in 3 of 10 strains tested (including type strain NCTC11637), separated the normally adjacent cagA and picA genes and did not involve IS605. Our results are discussed in terms of how cag-encoded proteins might help trigger the damaging inflammatory responses in the gastric epithelium and possible contributions of DNA rearrangements to genome evolution.

L13 ANSWER 17 OF 20 CAPLUS COPYRIGHT 2006 ACS on STN

1997:522873 Document No. 127:172134 The complete genome sequence of the gastric pathogen *Helicobacter pylori*. Tomb, Jean-F.; White, Owen; Kerlavage, Anthony R.; Clayton, Rebecca A.; Sutton, Granger G.; Fleischmann, Robert D.; Ketchum, Karen A.; Klenk, Hans Peter; Gill, Steven; Dougherty, Brian A.; Nelson, Karen; Quackenbush, John; Zhou, Lixin; Kirkness, Ewen F.; Peterson, Scott; Loftus, Brendan; Richardson, Delwood; Dodson, Robert; Khalak, Hanif G.; Glodek, Anna; McKenney, Keith; Fitzgerald, Lisa M.; Lee, Norman; Adams, Mark D.; Hickey, Erin K.; Berg, Douglas E.; Cocayne, Jeanine D.; Utterback, Teresa R.; Peterson, Jeremy D.; Kelley, Jenny M.; Cotton, Matthew D.; Weidman, Janice M.; Fujii, Claire; Bowman, Cheryl; Watthey, Larry; Wallin, Erik; Hayes, William S.; Borodovsky, Mark; Karp, Peter D.; Smith, Hamilton O.; Fraser, Claire M.; et al. (Inst. for Genomic Res., Rockville, MD, 20850, USA). *Nature* (London), 388(6642), 539-547 (English) 1997. CODEN: NATUAS. ISSN: 0028-0836. Publisher: Macmillan Magazines.

AB *Helicobacter pylori*, strain 26695, has a circular genome of 1,667,867 base pairs and 1590 predicted coding sequences. Sequence anal. indicates that *H. pylori* has well-developed systems for motility, for scavenging iron, and for DNA restriction and modification. Many putative adhesins, lipoproteins and other outer membrane proteins were identified, underscoring the potential complexity of host-pathogen interaction. Based on the large number of sequence-related genes encoding outer membrane proteins and the presence of homopolymeric tracts and dinucleotide repeats in coding sequences, *H. pylori*, like several other mucosal pathogens, probably uses recombination and slipped-strand mispairing within repeats as mechanisms for antigenic variation and adaptive evolution. Consistent with its restricted niche, *H. pylori* has a few regulatory networks, and a limited metabolic repertoire and biosynthetic capacity. Its survival in acid conditions depends, in part, on its ability to establish a pos. inside-membrane potential in low pH.

L13 ANSWER 18 OF 20 CAPLUS COPYRIGHT 2006 ACS on STN

1996:235631 Document No. 124:280413 Identification of murine helicobacters by PCR and restriction enzyme analyses. Riley, Lela K.; Franklin, Craig L.; Hook, Reuel R. Jr.; Besch-Williford, Cynthia (Department Veterinary Pathobiology, University Missouri, Columbia, MO, 65211, USA). *Journal of Clinical Microbiology*, 34(4), 942-6 (English) 1996. CODEN: JCMIDW. ISSN: 0095-1137. Publisher: American Society for Microbiology.

AB Three murine helicobacter species have recently been identified: *Helicobacter hepaticus*, *Helicobacter muridarum*, and *Helicobacter bilis*. Infections with *H. hepaticus* and *H. bilis* have been associated with hepatitis and hepatic neoplasia. In this study, oligonucleotide primers were designed from regions of the 16S rRNA gene that are conserved among members of the *Helicobacter* genus. The assay amplified the expected 374-bp product from all three rodent *Helicobacter* species and was able to detect as little as 5 pg of *H. hepaticus*, *H. bilis*, or *H. muridarum* DNA. The specificity of the reaction was determined by testing cecal DNA from uninfected mice and mice with documented *Helicobacter* infections and by testing DNA from other bacterial genera. A product of the expected size was generated with cecal DNA from *Helicobacter*-infected mice but not with DNA from uninfected mice. With the exception of that of "*Flexispira rappini*," which is closely related to the *Helicobacter* genus, DNA from other bacterial genera was not amplified with the *Helicobacter* genus-specific primers. MboI, MaeI, and HhaI restriction enzyme analyses of the amplified product were able to differentiate among the murine *Helicobacter* species but could not differentiate *H. bilis* from "*F. rappini*.". To distinguish *H. bilis*, a reverse primer based on *H. bilis* 16S rRNA sequence was designed. PCR with the *H. bilis*-specific reverse primer (Hbr) and the *Helicobacter* genus-specific forward primer (H276f) amplified *H. bilis* DNA but not DNA from "*F. rappini*" or other rodent helicobacters. Examination of a large number of murine cecal tissues with this combination of PCR assays and restriction enzyme analyses indicated that *H. hepaticus* and *H. bilis* infections are widespread in laboratory mouse and rat colonies.

L13 ANSWER 19 OF 20 CAPLUS COPYRIGHT 2006 ACS on STN

1995:547523 Document No. 123:107536 Long-term colonization with single and multiple strains of *Helicobacter pylori* assessed by DNA fingerprinting. Taylor, Nancy S.; Fox, James G.; Akopyants, Natalia S.; Berg, Douglas E.; Thompson, Nancy; Shames, Benjamin; Yan, Lili; Fontham, Elizabeth; Janney, Frances; et al. (Division Comparative Medicine, Massachusetts Institute Technology, Cambridge, MA, USA). *Journal of Clinical Microbiology*, 33(4), 918-23 (English) 1995. CODEN: JCMIDW. ISSN: 0095-1137.

AB The gastric pathogen *Helicobacter pylori* establishes long-term chronic infections that can lead to gastritis, peptic ulcers, and cancer. The species is so diverse that distinctly different strains are generally recovered from each patient. To better understand the dynamics of long-term carriage, the authors characterized *H. pylori* isolates from initial and follow-up biopsy specimens from a patient population at high risk of *H. pylori* infection and gastric cancer. Eighty-five isolates were obtained from 23 patients and were analyzed by genomic restriction enzyme anal., arbitrarily primed PCR fingerprinting, (random amplified polymorphic DNA anal.), and/or restriction of specific PCR-amplified genes (restriction fragment length polymorphism anal.). A single strain was found in sequential biopsy specimens from 12 of 15 patients (80%) receiving sucralfate. In the remaining three patients treated with sucralfate, two strains were identified in two patients and three strains were identified in the third patient. In contrast, a single strain was found in sequential biopsy specimens from only three of eight patients (37%) receiving bismuth, metronidazole, and nitrofurantoin. Two strains were identified in five other patients receiving bismuth-antibiotic (63%). IgG antibodies to *H. pylori* were present in the sera of all patients. Thus, *H. pylori* colonization can persist for long periods (up to at least 4 yr), despite high titers of IgG antibodies in serum. Resistance to metronidazole was noted in some strains before and/or after treatment, but all strains remained susceptible to amoxicillin, tetracycline, and nitrofurantoin. The authors conclude that *H. pylori* genotypes, as measured by several sensitive DNA fingerprinting methods, can remain stable for years in vivo, despite the acquisition or loss of drug resistance, circulating antibody, or exposure to antibiotics or sucralfate.

L13 ANSWER 20 OF 20 CAPLUS COPYRIGHT 2006 ACS on STN

1991:404368 Document No. 115:4368 Use of DNA restriction endonuclease digest and ribosomal RNA gene probe patterns to fingerprint *Helicobacter pylori* and *Helicobacter mustelae* isolated from human and animal hosts. Morgan, David D.; Owen, Robert J. (Cent. Public Health Lab., Natl. Collect. Type Cultures, London, NW9 5HT, UK). *Molecular and Cellular Probes*, 4(4), 321-34 (English) 1990. CODEN: MCPRE6. ISSN: 0890-8508.

AB Variation among strains of *H. pylori* and *H. mustelae* was examined by DNA restriction endonuclease digestion and rRNA gene patterns generated by using a nonradioactive probe. Chromosomal DNA was extracted from 30 cultures of *H. pylori* from human, Rhesus monkey, and pig gastric mucosa, and from 3 *H. mustelae* isolates from ferret gastric mucosa. DNA fingerprinting with HaeIII and HindIII showed *H. mustelae* was relatively homogeneous but revealed genomic heterogeneity within *H. pylori* with at least 18 different DNA patterns identifiable among the 30 isolates. Five sets of strains other than duplicates with matching DNA fingerprints were identified within *H. pylori*. The Peruvian isolates were the largest identical set and comprised 8 isolates from 4 different patients with 5 consecutive isolates from 1 patient. The Rhesus monkey strains were a relatively homogeneous set as were several Australian human isolates. The study demonstrates that rRNA gene restriction patterns provide a simple but highly discriminatory electrophoretic fingerprint for *H. pylori* with potential for use as a novel epidemiol. marker in addition to total DNA digest anal.

=> D L14 9,51 CBIB ABS

L14 ANSWER 9 OF 73 CAPLUS COPYRIGHT 2006 ACS on STN

2005:1004256 Document No. 143:301342 Cloning and sequence of type II restriction endonuclease HpyCII from *Helicobacter pylori* and potential genetic engineering application. Wang, Jin-town; Lin, Tzu-lung (Taiwan). U.S. Pat. Appl. Publ. US 2005202443 A1 20050915, 15 pp. (English). CODEN: USXXCO. APPLICATION: US 2004-796669 20040309.

AB Disclosed is a novel type II restriction endonuclease. More specifically, the nucleotide sequence and the encoded amino acid sequence of the restriction endonuclease HpyCII from *Helicobacter pylori* CCRC17132 are provided. Such enzyme recognizes a particular non-palindromic sequence of 5 oligonucleotides and cleaves DNA downstream of the DNA recognition sequence of nucleotides at the fourth base in the upper strand and the fifth base in the lower strand, and forms a one-base protruding end in the 5'-end after cleavage. The recognition and cleavage site of HpyCII is identical to the known restriction endonuclease BccI resp., (i.e. HpyCII is an isoschizomer of BccI), but the nucleotide sequence and the amino acid sequence are different from any other know restriction enzymes. The restriction endonuclease HpyCII can be used in the biotechnol. manipulation of genetic engineering and gene cloning.

L14 ANSWER 51 OF 73 CAPLUS COPYRIGHT 2006 ACS on STN

2001:851864 Document No. 136:66045 Characterization of type II restriction endonucleases (Hpy51-I) from *Helicobacter pylori* strain 51. Cho, Myung-Je; Park, Jeong-Uck; Jeon, Beong-Sam; Pack, Jeong-Won; Byun, Eun-Young; Lee, Sun-Kyung; Park, Ye-Hyoung; Song, Jae-Young; Lee, Woo-Kon; Baik, Seung-Chul; Choi, Yeo-Jeong; Jung, Seun-Ae; Choe, Mi-Young; Choi, Sang-Haeng; Ko, Gyung-Hyuck; Youn, Hee-Shang; Rhee, Kwang-Ho (Department of Microbiology, Pathology, and Pediatrics, Gyeongsang National University College of Medicine, Chunju, 660-280, S. Korea). Journal of Bacteriology and Virology, 31(3), 207-215 (Korean) 2001. CODEN: JBVOAH. ISSN: 1598-2467. Publisher: Journal of Bacteriology and Virology.

AB This study describes the purification and characterization of type II restriction endonuclease of *Helicobacter pylori* in order to understand the DNA restriction and modification of *H. pylori*. *H. pylori* cell extract was subjected to polyethyleneimine treatment salt precipitation, heparin-sepharose column chromatog., and fast protein liquid chromatog. (FPLC) using Resource Q column and Mono Q column to purify the type II restriction endonuclease. Hpy51-I was characterized to recognize the sequences 5'-GT(G/C)AC-3', yielding 5-base 5' protruding ends. The restriction sequence was identical to that of Tsp 45 I. The enzyme exhibited its maximal activity in the presence of 10-20 mM NaCl, but was inhibited completely in the presence of more than 80 mM NaCl. The enzyme showed its maximal activity in the presence of 1-10 mM MgCl₂. The optimal pH and temperature for enzyme activity was pH 9.0 and 37°C, resp. MnCl₂ could not substitute for MgCl₂ in reaction mixture. And addition of β-mercaptoethanol and bovine serum albumin in reaction mixture led to loss of enzyme activity of Hpy51-I. The whole cell extract of *H. pylori* strain 51 was confirmed to carry the enzyme activity for methylation of Hpy51-I-recognized sequence. Hpy51-I digested genomic DNAs of enteric bacteria to less than 1 kb while it could not cut the genomic DNAs of *H. pylori* isolates. In this study, the type II restriction enzyme (Hpy51-I) of *H. pylori* was identified and characterized, its biochem. properties demonstrating that Hpy51-I might be one of the barriers for preventing the introduction of foreign DNAs into *H. pylori*.

=> E WANG J/AU

=> S E35-E38

211 "WANG J L"/AU

9 "WANG J L F"/AU

1 "WANG J L H"/AU

1 "WANG J L S"/AU

L15 222 ("WANG J L"/AU OR "WANG J L F"/AU OR "WANG J L H"/AU OR "WANG J L S"/AU)

=> E WANG JIN/AU

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=> S E3,E130,E136
      946 "WANG JIN"/AU
      8 "WANG JIN T"/AU
      37 "WANG JIN TOWN"/AU
L16      991 ("WANG JIN"/AU OR "WANG JIN T"/AU OR "WANG JIN TOWN"/AU)

=> E LIN T/AU
=> S E3,E17
      135 "LIN T"/AU
      134 "LIN T L"/AU
L17      269 ("LIN T"/AU OR "LIN T L"/AU)

=> E LIN TZU/AU
=> S E3,E23
      9 "LIN TZU"/AU
      5 "LIN TZU LUNG"/AU
L18      14 ("LIN TZU"/AU OR "LIN TZU LUNG"/AU)

=> S L15,L16,L17,L18
L19      1491 (L15 OR L16 OR L17 OR L18)

=> S L13 AND L4
L20      20 L13 AND L4

=> S L20 NOT (L13,L14)
L21      0 L20 NOT ((L13 OR L14))
```

	L #	Hits	Search Text	DBs
1	L1	60483	RESTRICTION ADJ (ENDONUCLEASE OR ENZYME)	US- PGPUB; USPAT
2	L2	5517	HELICOBACTER	US- PGPUB; USPAT
3	L3	2334	L1 AND L2	US- PGPUB; USPAT
4	L4	33	L1 SAME L2	US- PGPUB; USPAT
5	L5	112	CCATC	US- PGPUB; USPAT
6	L6	111	GATGG	US- PGPUB; USPAT
7	L7	1	L4 AND (L5 OR L6)	US- PGPUB; USPAT
8	L8	10	L3 AND (L5 OR L6)	US- PGPUB; USPAT
9	L10	1	L7	US- PGPUB; USPAT
10	L9	42	L4 OR L8	US- PGPUB; USPAT